

Conservation Biology: a 'crisis discipline'

Fiona Hogan and Raylene Cooke

School of Life and Environmental Sciences, Deakin University, 221 Burwood Hwy, Burwood, Victoria, Australia 3125
email: fiona.hogan@deakin.edu.au

Abstract

Conserving biodiversity is of utmost importance on a global scale. Species conservation, however, is a challenging task, which is often compounded by a lack of knowledge of target species. New advances in information technology and molecular techniques, however, are enabling conservation biologists to obtain large amounts of data quickly, which will certainly aid in assigning conservation priorities. This article reviews the use of genetics in conservation biology and highlights, using the Powerful Owl *Ninox strenua* as an example, how DNA can be a valuable source of data. (*The Victorian Naturalist* 126 (3) 2008, 92-97)

Keywords: Powerful Owl, Conservation, Surrogate Species, DNA, Feathers

Uncontrolled manipulation of the world's ecosystems has resulted in the current rate of species loss being higher than at any previous time in human history (Soule 1991). This rapid decline in species richness, and the large number of species which are facing imminent extinction (Jetz *et al.* 2007), has been the trigger for the rapidly expanding field of conservation biology, also described as a 'crisis discipline' (Soule 1985).

Crisis disciplines arise from urgency, where there is an immediate need to understand the processes causing the crisis and to obtain knowledge on how to prevent, rectify or minimise its effects (DeSalle and Amato 2004). Consequently, there is a rapid expansion of tools used to solve these problems (Meine *et al.* 2006). Conservation biology is certainly one of these disciplines. It has benefited considerably from recent advances in both information technology and molecular biology techniques, which have enabled large amounts of data to be collected, stored and analysed. A large array of software packages designed specifically to interpret molecular data have become readily available (Lowe *et al.* 2004), and novel DNA markers provide a mechanism for understanding the ecology and biology of a diverse range of wildlife (DeYoung and Honeycutt 2005).

The integration of information technology and molecular techniques has accelerated the speed and accuracy of genetic analysis (DeSalle and Amato 2004). High-through put sequencing and multi-plex genotyping allows for a large number of genetic samples to be processed simultaneously (Bertorelle *et al.* 2004). Newer geospatial technologies such as geographic infor-

mation systems (GIS) are also being integrated with genetic analysis, which has given rise to yet another relatively new discipline, landscape genetics (Manel *et al.* 2003; Watts *et al.* 2004). The integration of these tools is enabling conservation biologists to collect data more rapidly, and provide improved management recommendations (DeYoung and Honeycutt 2005).

One of the major dilemmas that conservation biologists face is determining which species to conserve. It is impossible to monitor and manage every aspect of biodiversity and therefore, using a single species as a target is often adopted as a conservation tool (Simberloff 1998, Favreau *et al.* 2006). The ultimate aim of such an approach is to achieve community or ecosystem conservation by protecting a surrogate species. Surrogate species can have varying levels of ecological importance within ecosystems and therefore the identification of appropriate targets can deliver wider conservation goals (Wilcove 1993).

Keystone taxa are often selected as surrogate species (Simberloff 1998). These are species that have a critical ecological role in their ecosystem, where their disappearance has major implications beyond what might be expected, considering their biomass or abundance (Andelman and Fagan 2000). Identification of such species is beneficial for conservation, as their presence assures the ecological integrity of the communities they influence (Simberloff 1998). Keystone species, however, are difficult to identify without intricate knowledge of ecosystem dynamics, and there have been few detailed studies of keystone species (Simberloff 1998).

Surrogate species may also act as umbrellas, flagships or indicators. Umbrella species are often high-trophic-level mammalian or avian predators (Ozaki *et al.* 2006) which typically occupy large areas of habitat (Simberloff 1998). The protection of an umbrella species should theoretically save an entire suite of sympatric species with less demanding habitat requirements. Flagship species are those with high public appeal, usually large charismatic vertebrates, which are often used to promote environmental awareness (Simberloff 1998; Caro *et al.* 2004). The protection of flagship species and their habitat will lead to wider conservation benefits, where other species which share the same resources will inadvertently be protected (Andelman and Fagan 2000). Indicator species share some of the same habitat requirements as species, communities or ecosystems for which they indicate (Favreau *et al.* 2006) and therefore can be used to monitor ecosystem condition and health (Simberloff 1998). Although surrogate species are often employed by conservation biologists to help tackle conservation problems, the choice of particular surrogates is largely *ad hoc* (Landres *et al.* 1988). The use of surrogate species (umbrellas, flagships and indicators) has been found to have limited conservation benefits for protecting regional biota (Caro *et al.* 2004) and greater care in the choice of surrogate species may be required if they are to be successfully used in conservation biology (Caro and O'Doherty 1999).

Raptors as surrogate species

Humans have been fascinated with top-order predators such as raptors throughout history (Sergio *et al.* 2006). The charismatic appeal of these species has resulted in top-order predators being used as conservation targets for example, as flagship species to acquire financial support (White *et al.* 1997), raise environmental awareness (van Balen *et al.* 2000) and plan protected area systems (Andelman and Fagan 2000). Raptors are often perceived as highly sensitive with respect to their habitat and resource requirements, and therefore sensitive to habitat modification (Boal and Mannan 1999). They also have a low tolerance to disturbance (Thiollay 2006) where their breeding success, for example, is reduced by anthropogenic threats. Due to their vulnerability, raptors have been used as indicator species, where their presence can be an indica-

tion of a particular habitat quality (Sergio *et al.* 2006).

Owls are often used as flagship species for conservation campaigns, probably none more so than the Northern Spotted Owl *Strix occidentalis caurina* in the United States of America (Simberloff 1998). The mandatory requirement for the US Forest Service to use management surrogate species led to the Northern Spotted Owl becoming the flagship for the Pacific Northwest Region of the USA (Dunk *et al.* 2006). The rationale for the decision was three fold: (1) it was a threatened species; (2) it was charismatic; and (3) it was reliant on large amounts of old-growth forest (Lamberson *et al.* 1992). As the Northern Spotted Owl requires large areas of old growth forest for its survival and reproduction, it was assumed that many other species, which also rely on old growth forest, would retrospectively be protected (Lamberson *et al.* 1994). The conservation of the Northern Spotted Owl would therefore serve not only as a flagship species but also as an umbrella species.

Similarly, in Australia the Powerful Owl *Ninox strenua* has also been used as a surrogate species, particularly in regard to forestry operations and urban planning (Loyn *et al.* 2001). To help protect the Powerful Owl from the adverse impacts of timber harvesting, large amounts of forest have been reserved in Powerful Owl management areas (POMAs) to provide sufficient habitat for the owl (McCarthy *et al.* 1999). The conservation of this habitat should also protect sympatric species within these forested environments; so the Powerful Owl is serving as an umbrella species within these ecosystems.

The charismatic appeal of the Powerful Owl has led to its high public profile. It is often used as a flagship species for urban development. A recent example was in the development of a major freeway extension in Melbourne's outer eastern suburbs. The presence of the Powerful Owl in the Mullum Mullum creek corridor was used by conservationists to rally against the proposed new freeway developments. Media releases stated that '... the Powerful Owl lived up to its name in the eastern suburbs' because the government committed \$326 million to an alternative freeway route, including a 1.5 km tunnel under the Mullum Mullum Creek corridor to protect habitat for the Powerful Owl (Tinkler 2004). While original flora and fauna



Female Powerful Owl *Ninox strenua*. Photo by Fiona Hogan



Fig. 1. The distribution of the Powerful Owl in Australia (adapted from Higgins 1999)

surveys of the Mullum Mullum creek corridor revealed that the proposed area of construction contained high intrinsic habitat value and that effects of the proposed development on flora and fauna would be considerable (Department of Conservation 1990), this report made no specific mention of the Powerful Owl. Regardless of this, the Powerful Owl was still used as a flagship species in this instance, and implementation of the report was successful in preserving valuable habitat within the urban matrix of Melbourne.

The Powerful Owl is the largest and arguably the most charismatic owl species in Australia. It is endemic to Australia and is distributed across the three eastern mainland states: Victoria, New South Wales and Queensland (Fig. 1). It has a limited distribution along the east coast (Garnett and Crowley 2000), where human population density and urban growth are also particularly high (Luck 2007).

The Powerful Owl is of international concern and is listed in Appendix II of CITES (Convention on International Trade of Endangered Species of Wild Fauna and Flora) and considered

Least Concern by the IUCN (2001 IUCN Red List of Threatened Species). Nationally, it is classified as of conservation significance (Higgins 1999) and vulnerable within the States of Victoria (Department of Sustainability and Environment 2003), New South Wales (Olsen 1998) and Queensland (Olsen 1998).

Risks to the persistence of Powerful Owls pertain largely to the loss or degradation of essential habitat (Brouwer and Garnett 1990). Traditionally perceived as a habitat and dietary specialist, the Powerful Owl was thought to require continuous tracts of old growth forest (Schodde and Mason 1980; Debus and Chafer 1994). The large body size and high metabolic rate of Powerful Owls necessitates a large, energy-rich diet, comprising medium-sized arboreal prey such as possums (Webster *et al.* 1999; Cooke *et al.* 2002a) and successful reproduction requires the presence of tree hollows suitable for nesting (Cooke *et al.* 2002b). Current research, however, suggests that the Powerful Owl is more adaptable than once perceived, inhabiting forest and woodland remnants close to major urban centres, including Brisbane, Melbourne

and Sydney (Fig. 1) (Pavey 1993; Cooke *et al.* 2002a; Kavanagh 2004). It is uncertain whether owls in these urban centres are remnant populations (Kavanagh 2004) or are associated with the abundance of potential prey species and the increased protection of remnant patches within the urban matrix (Cooke *et al.* 2006).

The presence of Powerful Owls in major Australian cities is of importance, especially considering the conservation significance of this species. Surveys conducted by Miller (2003) indicated that Victorians have a relatively strong emotional attachment to individual animals and are interested in learning about wildlife and the natural environment. The presence of the Powerful Owl in urban environments, therefore, provides the opportunity to use the charismatic appeal of this species to promote environmental awareness amongst city people, and as a surrogate species for conservation. However, in order to successfully use the Powerful Owl as an umbrella, flagship or indicator species, further knowledge about its ecology, biology and habitat requirements is fundamental.

Although the Powerful Owl has been the focus of numerous studies over the past 30 years, knowledge on many aspects of its ecology and biology remain unknown. Most published studies have focused on diet (e.g. Chafer 1992; Debus and Chafer 1994; Cooke *et al.* 1997; Webster *et al.* 1999; Cooke *et al.* 2002a; Cooke *et al.* 2006) and habitat preference (e.g. Schodde and Mason 1980; Debus and Chafer 1994; Kavanagh 1998; Cooke *et al.* 2002b). Critical data on other aspects of Powerful Owl biology and ecology, such as mating systems, population structure and dispersal are currently unknown. Their dispersed distribution, low population densities, nocturnal activity cycle and difficulty in establishing the identity of individual birds has inhibited the collection of this information.

The use of genetics in conservation biology

Conservation genetics presents an opportunity to reduce current knowledge gaps in ecological studies. Genetic information can strengthen conservation knowledge and ensure that rational management decisions are made (DeSalle and Amato 2004). Microsatellites (short tandem nucleotide repeats) have become the genetic marker of choice for studies of intraspecific variation of wild populations (DeWoody

2005). A limitation of this approach is that microsatellite markers usually have to be developed for the species under investigation (Sunnucks 2000) which can be time-consuming and costly (Piggott and Taylor 2003). Once developed, however, microsatellite markers can be used in other closely related species, so that the development process does not need to be repeated for every species (Piggott and Taylor 2003).

A number of microsatellite markers used in combination, can provide a DNA profile which can unequivocally identify individuals (Piggott and Taylor 2003). Hogan *et al.* (2007) developed a suite of 14 microsatellite DNA markers from the Powerful Owl. The resolution of these markers was sufficient to provide a probability of identity (P_{ID}) of 0.0001 (1 in 10 000) for unrelated Powerful Owls, which is more than sufficient for a species with a relatively small population size (~7,000 breeding adults) and sparse distribution (Garnett and Crowley 2000). Individual DNA profiling allows for crucial elements of the breeding ecology to be assessed and the relatedness of individuals to be determined which can identify inbreeding (Galeotti *et al.* 1997). When gender is inferred by employing sex-specific markers, DNA profiling can be an extremely valuable tool, for identifying putative parents, inferring mating systems, assessing sex bias dispersal and sex-ratios of offspring.

Genetic analysis traditionally required large amounts of DNA, therefore, studies involving wild animals employed destructive sampling where the animal was killed to obtain tissue samples (Taberlet *et al.* 1999). An alternative was to capture the animal to obtain tissue samples without killing it, but this is traumatic and accidental death can occur. These two types of sampling have the advantage of providing abundant good quality DNA (Taberlet *et al.* 1999); however, neither are conducive to conservation biology. Another disadvantage of the non-destructive sampling is that capture may alter the normal behaviour of the individual being studied (Morin *et al.* 1994).

Development of the polymerase chain reaction (PCR) in the early 1980's (Saiki *et al.* 1985) enabled very small amounts of sample to be used for genetic analysis. PCR involves DNA sequence being amplified or 'copied' by enzymatic reaction *in vitro*, using short pieces of DNA (primers). Millions of copies of the

target DNA sequence can be produced, which can subsequently be used for a range of genetic analyses. PCR enables the implementation of non-destructive and non-invasive sampling techniques, where DNA can be obtained from small amounts of tissue (biopsy samples), blood, feathers, hair or trace material left by the animal. PCR has therefore provided the greatest breakthrough in terms of genetics in conservation biology, as it has eliminated the need to destroy animals for research.

PCR has made possible the alternative sampling technique of non-invasive genetic sampling (NGS), where DNA left behind by an animal, such as shed hair, scats and feathers can be collected (Waits and Paetkau 2005). The attractiveness of this technique is the opportunity to obtain genetic material from free-ranging animals without having to catch, handle or even observe them (Taberlet and Luikart 1999). This technique is especially valuable when studying species that are rare, endangered or cryptic (Piggott and Taylor 2003), where using invasive study methods such as trapping is neither feasible nor appropriate (Greenwood 1996).

Shed feathers are a readily available DNA source from species which regularly moult, such as the Powerful Owl. Feathers can be collected from underneath roosts and easily identified through comparison to museum specimens. Hogan *et al.* (2008) demonstrated that a large number of DNA samples (shed feathers) can be collected over a large spatial scale, within a relatively short period of time. This mode of DNA sampling is revolutionary for ecological studies, and will provide data which otherwise would be impossible to obtain through traditional ecological techniques such as banding. The analysis of DNA extracted from samples can provide a wealth of information such as individual identification, estimates of relatedness, pedigree reconstruction, sex identification, estimates of census and effective population size, and the level of genetic polymorphism within or between populations (Taberlet and Luikart 1999; Piggott and Taylor 2003). Information obtained from such genetic data will greatly improve our knowledge about the biology and ecology of species, such as the Powerful Owl, which can further be disseminated into management strategies and subsequently enhance the effectiveness of future conservation efforts.

References

- Andelman SJ and Fagan WF (2000) Umbrellas and flagships: efficient conservation surrogates or expensive mistakes? *Proceedings of the National Academy of Sciences of the United States of America* **97**, 5954-5959.
- Bertorelle G, Bruford M, Chemini C, Vernesi C and Hauffe HC (2004) New, flexible Bayesian approaches to revolutionize conservation genetics. *Conservation Biology* **18**, 584-584.
- Boal CW and Mannan RW (1999) Comparative breeding ecology of Cooper's Hawks in urban and exurban areas of southeastern Arizona. *Journal of Wildlife Management* **63**, 77-84.
- Brouwer J and Garnett S (1990) *Threatened birds of Australia - an annotated list*.
- Caro T, Engilis A, Fitzherbert F and Gardner T (2004) Preliminary assessment of the flagship species concept at a small scale. *Animal Conservation* **7**, 63-70.
- Caro T and O'Doherty G (1999). On the use of surrogate species in conservation biology. *Conservation Biology* **13**, 805-814.
- Chafer C (1992) Observations of the Powerful Owl *Ninox strenua* in the Illawarra and Shoalhaven Regions of New South Wales. *Australian Bird Watcher* **14**, 289-300.
- Cooke R, Wallis R, Webster A and Wilson J (1997) Diet of a family of Powerful Owls *Ninox strenua* from Warrandyte, Victoria. *Proceedings of the Royal Society of Victoria* **109**, 1-6.
- Cooke R, Wallis R and Webster A (2002a) Urbanisation and the Ecology of Powerful Owls *Ninox strenua* in outer Melbourne, Victoria. In *Ecology and Conservation of Owls*, pp. 100-106. Eds I Newton, R Kavanagh, P Olsen, and I Taylor. (CSIRO: Melbourne, Australia).
- Cooke R, Wallis R and White J (2002b) Use of vegetative structure by Powerful Owls in outer urban Melbourne, Victoria, Australia - Implications for management. *Journal of Raptor Research* **36**, 294-299.
- Cooke R, Wallis R, Hogan F, White J and Webster A (2006). The diet of Powerful Owls *Ninox strenua* and prey availability in a continuum of habitats from disturbed urban fringe to protected forest environments in south-eastern Australia. *Wildlife Research* **33**, 199-206.
- Debus S and Chafer C (1994) The Powerful Owl *Ninox strenua* in New South Wales. *Australian Birds* **28**, S21-S64.
- Department of Conservation F. a. L. (1990) Flora and fauna of the Koonung and Mullum Mullum Valleys (Proposed Eastern arterial road and Ringwood bypass) Victoria. Department of Conservation, Forests and Lands, Melbourne, Australia.
- Department of Sustainability and Environment (2003) Advisory list of threatened vertebrate fauna in Victoria. Department of Sustainability and Environment, Melbourne.
- DeSalle R and Amato G (2004) The expansion of conservation genetics. *Nature Reviews Genetics* **5**, 702-712.
- DeWoody JA (2005) Molecular approaches to the study of parentage, relatedness, and fitness: Practical applications for wild animals. *Journal of Wildlife Management* **69**, 1400-1418.
- DeYoung RW and Honeycutt RL (2005) The molecular toolbox: Genetic techniques in wildlife ecology and management. *Journal of Wildlife Management* **69**, 1362-1384.
- Dunk JR, Zielinski WJ and Welsh HH (2006) Evaluating reserves for species richness and representation in northern California. *Diversity and Distributions* **12**, 434-442.
- Favreau JM, Drew CA, Hess GR, Rubino MJ, Koch FH and Eschelbach KA (2006) Recommendations for assessing the effectiveness of surrogate species approaches. *Biodiversity and Conservation* **15**, 3949-3969.
- Galeotti P, Pilastro A, Tavecchia G, Bonetti A and Congiu L (1997) Genetic similarity in Long-eared Owl communal winter roosts: a DNA fingerprinting study. *Molecular Ecology* **6**, 429-435.

- Garnett ST and Crowley GM (2000) *The Action Plan for Australian Birds*. (Environment Australia: Canberra)
- Greenwood J (1996) Basic techniques. In *Ecological Census Techniques, A Handbook* pp. 11-110. Ed W. Sutherland. (Cambridge University Press: Cambridge).
- Higgins PJE (1999) *Handbook of Australian, New Zealand and Antarctic Birds. Volume 4: Parrots and Dollarbird*. (Oxford University Press: Melbourne)
- Hogan F, Burrige C, Cooke R and Norman J (2007) Isolation and characterisation of microsatellite loci to DNA fingerprint the Powerful Owl *Ninox strenua*. *Molecular Ecology Notes* 7, 1305 - 1307.
- Hogan F, Cooke R, Burrige C and Norman J (2008) Optimizing the use of shed feathers for genetic analysis. *Molecular Ecology Resources* 8, 561-567.
- Jetz W, Wilcove DS and Dobson AP (2007) Projected impacts of climate and land-use change on the global diversity of birds. *PLoS Biology* 5, 1211-1219.
- Kavanagh RP (1998) Ecology and management of large forest owls in southeastern Australia. *Australian Journal of Ecology* 23, 184-185.
- Kavanagh RP (2004) Conserving owls in Sydney's urban bushland: current status and requirements. *Urban Wildlife* 93-108.
- Lamberson RH, McKelvey R, Noon BR and Voss C (1992) A dynamic analysis of Northern Spotted Owl viability in a fragmented forest landscape. *Conservation Biology* 6, 505-512.
- Lamberson RH, Noon BR, Voss C and McKelvey KS (1994) Reserve design for territorial species - the effects of patch size and spacing on the viability of the Northern Spotted Owl. *Conservation Biology* 8, 185-195.
- Landres PB, Verner J and Thomas JW (1988) Ecological uses of vertebrate indicator species - a critique. *Conservation Biology* 2, 316-328.
- Lowe A, Harris S and Ashton P (2004) *Ecological Genetics: Design, Analysis and Application*. (Blackwell Publishing, Victoria, Australia).
- Loyn RH, McNabb EG, Volodina L and Willig R (2001) Modelling landscape distributions of large forest owls as applied to managing forests in north-east Victoria, Australia. *Conservation Biology* 97, 361-376.
- Luck GW (2007) The relationships between net primary productivity, human population density and species conservation. *Journal of Biogeography* 34, 201-212.
- Manel S, Schwartz MK, Luikart G and Taberlet P (2003) Landscape genetics: combining landscape ecology and population genetics. *Trends in Ecology and Evolution* 18, 189-197.
- McCarthy M A, Webster A, Loyn RH and Lowe KW (1999) Uncertainty in assessing the viability of the Powerful Owl *Ninox strenua* in Victoria, Australia. *Pacific Conservation Biology* 5, 144-154.
- Meine C, Soule M and Noss RF (2006) "A mission-driven discipline": the growth of conservation biology. *Conservation Biology* 20, 631-651.
- Miller KK (2003) Public and stakeholder values of wildlife in Victoria, Australia. *Wildlife Research* 30, 465-476.
- Morin PA, Wallis J, Moore JJ and Woodruff DS (1994) Paternity exclusion in a community of wild chimpanzees using hypervariable simple sequence repeats. *Molecular Ecology* 3, 469-477.
- Olsen P (1998) *Australia's Raptors: Diurnal Birds of Prey*. (Birds Australia: Melbourne).
- Ozaki K, Isono M, Kawahara T, Iida S, Kudo T and Fukuyama K (2006) A mechanistic approach to evaluation of umbrella species as conservation surrogates. *Conservation Biology* 20, 1507-1515.
- Pavey CR (1993) The Distribution and Conservation Status of the Powerful Owl (*Ninox strenua*) in Queensland. In *Australian Raptor Studies*, pp. 144-153. Ed P Olsen (Australian Raptor Association Melbourne: Australia).
- Piggott MP and Taylor AC (2003) Remote collection of animal DNA and its applications in conservation management and understanding the population biology of rare and cryptic species. *Wildlife Research* 30, 1-13.
- Saiki RK, Scharf S, Faloona F, Mullis KB, Horn GT, Erlich HA and Arnheim N (1985) Enzymatic amplification of beta-globin genomic sequences and restriction site analysis for diagnosis of sickle-cell anemia. *Science* 230, 1350-1354.
- Schodde R and Mason IJ (1980) *Nocturnal Birds of Australia* (Lansdowne Editions: Melbourne)
- Sergio F, Newton J, Marchesi L and Pedrini P (2006). Ecologically justified charisma: preservation of top predators delivers biodiversity conservation. *Journal of Applied Ecology* 43, 1049-1055.
- Simberloff D (1998) Flagships, umbrellas, and keystones: is single-species management passe in the landscape era? *Biological Conservation* 83, 247-257.
- Soule ME (1985) What is conservation biology? *Bioscience* 35, 727-734.
- Soule ME (1991) Conservation - tactics for a constant crisis. *Science* 253, 744-750.
- Sunnucks P (2000) Efficient genetic markers for population biology. *Trends in Ecology and Evolution* 15, 199-203.
- Taberlet P and Luikart G (1999) Non-invasive genetic sampling and individual identification. *Biological Journal of the Linnean Society* 68, 41-55.
- Taberlet P, Waits LP and Luikart G (1999) Noninvasive genetic sampling: look before you leap. *Trends in Ecology and Evolution* 14, 323-327.
- Thiollay JM (2006) The decline of raptors in West Africa: long-term assessment and the role of protected areas. *Ibis* 148, 240-254.
- Tinkler C (2004) \$400m bill for creature comfort. *Sunday Herald Sun*, Melbourne. 29
- van Balen S, Nijman V and Prins HHT (2000) The Javan hawk-eagle: misconceptions about rareness and threat. *Biological Conservation* 96, 297-304.
- Waits LP and Paetkau D (2005) Noninvasive genetic sampling tools for wildlife biologists: a review of applications and recommendations for accurate data collection. *Journal of Wildlife Management* 69, 1419-1433.
- Watts P C, Rouquette JR, Saccheri J, Kemp SJ and Thompson DJ (2004) Molecular and ecological evidence for small-scale isolation by distance in an endangered damselfly, (*Coenagrion mercuriale*). *Molecular Ecology* 13, 2931-2945.
- Webster A, Cooke R, Jameson G and Wallis R (1999) Diet, roosts and breeding of Powerful Owls *Ninox strenua* in a disturbed, urban environment: a case for cannibalism? Or a case of infanticide? *Emu* 99, 80-83.
- White PCL, Gregory KW, Lindley PJ and Richards G (1997) Economic values of threatened mammals in Britain: a case study of the Otter *Lutra lutra* and the Water Vole *Arvicola terrestris*. *Biological Conservation* 82, 345-354.
- Wilcove D (1993) Getting ahead of the extinction curve. *Ecological Applications* 3, 218-220.

Received 3 November 2008; accepted 17 May 2009